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Research Note

Comparison of Weep and Carcass Rinses for Recovery of *Campylobacter* from Retail Broiler Carcasses

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ABSTRACT

Campylobacter is frequently recovered from broiler carcasses. Carcass rinsing is a commonly used procedure for isolating *Campylobacter* from poultry. A viscous fluid, or weep, exudes from broiler carcasses that have been packaged. This fluid can contain bacteria that were attached to the carcass and represents a potential means of detecting *Campylobacter*-contaminated carcasses through cultural analysis. Experiments were conducted to compare the efficacy of a weep sampling method with that of a carcass rinse method. For both trials, retail carcasses were purchased. Packages were opened, and 0.1-ml aliquots of weep fluid from the retail packages were plated onto Campy-cefex agar. Carcasses were removed from the package and rinsed in 100 ml of sterile water. Next, 0.1-ml aliquots of the rinsate were plated onto Campy-cefex agar and incubated. In a second experiment, samples were both directly plated and enriched in Bolton enrichment broth. In the first experiment, 35 of 60 carcass rinses tested positive for *Campylobacter,* while 29 of 60 weep samples yielded *Campylobacter* isolates with levels of 1.0 and 1.1 log CFU/ml, respectively. In the second experiment, *Campylobacter* was recovered from 9 of 40 rinse samples and from 13 of 40 weep samples by direct plating, while the organism was recovered from 28 of 40 rinses samples and from 23 of 40 carcass samples by enrichment. There was no significant difference between the two methods with respect to *Campylobacter* prevalence as determined by the chi-square test. *Campylobacter* levels recovered by both methods averaged 0.9 log CFU/ml. The sampling of weep fluid was a simple, effective means of detecting this important human enteropathogen on broiler carcasses.

Campylobacteriosis is a leading cause of foodborne disease worldwide. *Campylobacter,* the bacterial species that causes this disease, is frequently isolated from broiler carcasses, even from retail sources. As the food safety relevance of this organism has increased, so has the need for efficient, effective methods for its detection and enumeration in samples from poultry products *(4, 6).*

A common means of recovering this organism from broiler carcasses is a carcass rinse procedure. A standard carcass sampling procedure used by the U.S. Department of Agriculture's Food Safety and Inspection Service (USDA-FSIS) requires the carcass to be rinsed in approximately 200 ml of buffered peptone water *(13).* An alternative procedure requiring only 100 ml decreases medium volume and incubator space and increases the bacterial cell–to–liquid ratio without decreasing the recovery rate *(5, 18).* The same volume of sterile water has successfully been employed to recover *Campylobacter* from broiler carcasses *(12, 17, 18).*

A viscous fluid, or weep, exudes from poultry products that have been packaged for retail display. This material has been analyzed culturally for the isolation and enumeration of bacteria. Mercuri and Kotula *(10)* found that the culturing of this material compared favorably with a swab technique for the enumeration of aerobic bacteria from broiler carcasses. Clark and Bueschkens *(3)* recovered *Campylobacter* from retail chicken parts by direct plating and enrichment of the ''exuded blood and water.'' The purpose of the present study was to compare the efficacy of cultural isolation from carcass rinse samples and with that of cultural isolation from carcass drip, or weep, samples for the detection of *Campylobacter* in poultry carcasses.

MATERIALS AND METHODS

For the first experiment, 60 whole broiler carcasses were purchased from local retail sources and transported to the laboratory. Six replicate experiments with 10 carcasses, each of which was processed on the same day and by the same processor, were performed. Each package was sanitized with 70% ethanol and opened by cutting one end with sterile scissors. Each carcass was removed aseptically and placed in a bag with 100 ml of sterile water. Each bagged whole carcass was then shaken vigorously for 1 min. After each carcass was rinsed, 0.1 ml of rinsate was plated onto duplicate Campy-cefex agar plates *(19).* Once the carcass was removed from the retail carcass bag, 0.1 ml of weep was plated onto duplicate Campy-cefex agar plates. *Weep* was defined as the bloody fluid that remained in the bag once the carcass was removed.

Plates were then incubated for 36 to 48 h at 42°C in a microaerobic atmosphere (5% O_2, 10% CO_2, and 85% N_2). After incubation, plates were examined for the presence of presumptive

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TABLE 1. *Prevalence rates for and levels of* Campylobacter *as determined by plating carcass weep fluid and carcass rinsate onto Campy-cefex agar*

	Campylobacter count (log_{10} CFU/ml) for sample type		Prevalence for sample type[a]		
Replicate	Weep	Rinse	Weep	Rinse	Either type
1	0.0	0.0	0/10	0/10	0/10
2	1.0	0.8	6/10	6/10	7/10
3	1.6	1.4	8/10	9/10	9/10
4	0.9	1.0	7/10	9/10	9/10
5	1.4	1.1	5/10	7/10	7/10
6	0.8	0.8	3/10	4/10	7/10
Total	1.1	1.0	29/60	35/60	39/60

[a] Number of samples positive/number of samples tested.

Campylobacter colonies. Presumptive colonies are flat or raised, round, entire, cream or tan colored, and smooth or glistening and are translucent when the petri plate is held in front of a light source. Representative presumptive colonies were confirmed by the observation of spiral cells and rapid, darting motility in wet mount preparations with the use of phase-contrast microscopy. Counts from plates with confirmed *Campylobacter* colonies were converted to log_{10} values and recorded. Prevalence rates for each sample type and method were analyzed with a chi-square test for independence *(16).*

For the second experiment, 40 retail broiler carcasses were sampled in four replicate trials as described for the first experiment. However, aliquots of rinsate and weep were enriched in Bolton broth (Oxoid) in addition to being directly plated as described above. A 1-ml aliquot of sample (rinse or weep) was added to 100 ml of Bolton enrichment broth in a Ziploc bag. Enrichments were incubated at 42°C for 36 to 48 h in a microaerobic atmosphere before being plated onto Campy-cefex agar. After a sample was evaluated, confirmed, and analyzed as described above, the result for that sample (positive or negative for *Campylobacter*) was recorded.

RESULTS

Prevalence rates and levels (log_{10} CFU/ml of weep or rinse) of *Campylobacter* determined by plating on Campy-cefex agar in the first experiment are reported in Table 1. Forty-eight percent (29 of 60) of the directly plated weep samples tested positive for *Campylobacter,* compared with 58% (35 of 60) of the rinse samples; the difference was not significant by the chi-square test for independence *(16).* Analysis of either sample type revealed that 65% of the retail samples were *Campylobacter* positive. Average levels for each sample type differed by only 0.1 log_{10} CFU/ml of sample.

Results for the second experiment are reported in Tables 2 and 3. Thirty-two percent of the directly plated weep samples tested positive for *Campylobacter,* compared with only 22% of the directly plated rinse samples (Table 2); the difference was not significant by the chi-square test for independence *(16).* Average population levels for each sample type were identical (Table 3). After enrichment, *Campylobacter* prevalence increased to 57 and 70% for weep and rinse samples, respectively. When either method (direct plating or enrichment) or sample type (weep or rinse) was considered, 90% (36/40) of the samples tested positive for the organism.

DISCUSSION

Since the genus was first proposed in 1967, *Campylobacter* has been established as one of the most significant bacterial foodborne pathogens in developed countries worldwide. A risk factor for campylobacteriosis is the consumption of undercooked poultry *(4, 6). Campylobacter* is frequently isolated from poultry and poultry products, with >90% of samples testing positive for the organism in some studies *(9, 11, 12, 15, 17).*

While cultural methods for the analysis of foods have improved over the years, the development of such methods

TABLE 2. *Prevalence of* Campylobacter *as determined by enrichment of weep fluid and carcass rinsate in Bolton enrichment broth*[a]

	Prevalence by direct plating of sample type		Prevalence by enrichment of sample type		
Replicate	Weep	Rinse	Weep	Rinse	Prevalence for either method or sample type
1	2/10	2/10	2/10	8/10	8/10
2	2/10	3/10	5/10	6/10	7/10
3	5/10	3/10	10/10	7/10	10/10
4	4/10	1/10	6/10	7/10	9/10
Total	13/40	9/40	23/40	28/40	36/40

[a] Prevalence is expressed as number of samples positive/number of samples tested.

TABLE 3. *Levels of* Campylobacter *as determined by directly plating carcass weep fluid and carcass rinsate onto Campy-cefex agar* (n = *40)*

Replicate	*Campylobacter* county (log_{10} CFU/ml) for sample type	
	Weep	Rinse
1	0.7	0.7
2	0.8	1.2
3	1.2	1.1
4	0.9	0.9
Average	0.9	0.9

continues. There are a number of approaches that can be taken to isolate *Campylobacter* from poultry products. These approaches include direct plating and enrichment of neck skin, swabs, and rinses and even incubation of the entire product in enrichment broth *(1, 2, 5, 7–9, 12, 14, 15, 17).* However, carcass rinses are most often used by regulatory agencies. The USDA-FSIS employs a standard method that involves the rinsing of carcasses with 200 ml of dilute buffered peptone water *(13).* However, *Campylobacter* can be consistently recovered from carcasses by rinsing with as little as 100 ml of sterile water *(5, 18).*

Previously published literature indicates that bacteria can be harvested from the "drip" or "exuded blood and water" of packaged poultry carcasses or parts *(10).* Clark and Bueschkens *(3)* successfully isolated *Campylobacter* from this fluid. The present study was undertaken to determine whether weep fluid from retail broiler carcasses would yield *Campylobacter* as consistently as carcass rinse sampling did.

While the direct plating of fresh carcass rinses often yields prevalence rates of >90%, lower prevalence rates are frequently reported for retail products *(2, 17).* In the present study, prevalence rates for directly plated retail samples, whether these samples were weep or carcass rinse samples, were similar to those previously reported. Rogol et al. *(14)* detected *Campylobacter* in 37% of 70 chicken meat samples, whereas Stern and Line *(17)* found only 20% of retail carcasses to be *Campylobacter* positive.

No other reports of *Campylobacter* levels in weep samples were noted in searches of published literature. Levels reported for both weep and carcass rinse samples were similar to those previously reported for carcass rinses. Line et al. *(9)* reported *Campylobacter* levels of 2.2 log CFU/ml of rinse for freshly processed broilers. Stern and Robach *(18)* reported *Campylobacter* levels of slightly more than 1.0 log CFU/ml of rinse from fresh carcasses.

The enrichment of both weep and carcass rinse samples resulted in increased *Campylobacter* detection. Aquino et al. *(2)* reported a decrease in the detection of *Campylobacter* in fresh carcasses with enrichment (from 95 to 48%), but Stern and Line *(17)* reported a *Campylobacter* recovery rate of 95% with enrichment, compared with 20% by direct plating. Smeltzer *(15)* reported an increase in detection (from 84 to 94%) with enrichment. In the present study, maximum detection was achieved when both direct plating and enrichment of both sample types were considered. Similar trends have previously been reported. Jorgensen et al. *(7)* and Kramer et al. *(8)* found >80% of poultry samples to be *Campylobacter* positive when a variety of methods were used. No single method or medium type developed to date is entirely effective in recovering this fastidious organism from poultry meat. This point is demonstrated by the results of the present study, in which a number of carcasses tested positive for *Campylobacter* by only one of the methods (direct plating or enrichment) or for only one of the sample types (weep or carcass rinse).

Weep samples analyzed in this study were obtained more easily and quickly than rinse samples. *Campylobacter* levels obtained for both sample types were nearly identical. There were no statistical differences between recovery rates for the two sample types whether direct plating or enrichment was employed. The use of weep samples for the detection and enumeration of *Campylobacter* may be a suitable alternative to the use of carcass rinse samples, particularly when materials or technical help are in short supply.

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